PATENT COOPERATION TREATY

PCT

INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	**FOR FURTHER ACTION**	see Form PCT/ISA/220 as well as, where applicable, item 5 below.
BCS03-2007		
International application No.	International filing date *(day/month/year)*	(Earliest) Priority Date *(day/month/year)*
PCT/EP2004/013122	17/11/2004	18/11/2003
Applicant		
BAYER BIOSCIENCE N.V.		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 10 sheets.

[X] It is also accompanied by a copy of each prior art document cited in this report.

1. **Basis of the report**
   a. With regard to the **language,** the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.
      [ ] The international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).
   b. [X] With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, see Box No. I.

2. [ ] **Certain claims were found unsearchable** (See Box II).

3. [X] **Unity of invention is lacking** (see Box III).

4. With regard to the **title,**
   [X] the text is approved as submitted by the applicant.
   [ ] the text has been established by this Authority to read as follows:

5. With regard to the **abstract,**
   [X] the text is approved as submitted by the applicant.
   [ ] the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box No. IV. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. With regard to the **drawings,**
   a. the figure of the **drawings** to be published with the abstract is Figure No. 1
      [X] as suggested by the applicant.
      [ ] as selected by this Authority, because the applicant failed to suggest a figure.
      [ ] as selected by this Authority, because this figure better characterizes the invention.
   b. [ ] none of the figures is to be published with the abstract.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP2004/013122

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.b of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, the international search was carried out on the basis of:

   a. type of material

   [X] a sequence listing

   [ ] table(s) related to the sequence listing

   b. format of material

   [X] in written format

   [X] in computer readable form

   c. time of filing/furnishing

   [X] contained in the international application as filed

   [X] filed together with the international application in computer readable form

   [ ] furnished subsequently to this Authority for the purpose of search

2. [ ] In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP2004/013122

Box II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [ ] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. [X] Claims Nos.: 24-26
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

see FURTHER INFORMATION sheet PCT/ISA/210

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. [X] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[X] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box II.2

Claims Nos.: 24-26

Present claims 24 to 26 relate to a nucleic acid defined inter alia by the sequence of the encoded protein and by reference to the parameter of the GC content of about 50% to about 60%.
In addition, said claims use provisos, i. e. negative features, to define their subject matter. In the case of claim 25, 100 provisos are recited, which make it unduly burdensome to determine the scope of protection sought. The large amount of provisions and conditions in said claims results in a lack of conciseness within the meaning of Article 6 PCT.
The use of the above-mentioned parameter and the use of predominantly negative features in the present context, which make it difficult to determine the boundaries of the claimed invention, is considered to lead to a lack of clarity within the meaning of Article 6 PCT. It is impossible to compare the parameters and the multitude of provisos the applicant has chosen to employ with what is set out in the prior art. The lack of clarity is such as to render a meaningful search impossible.

Consequently, no search report has been established for the subject matter of present claims 24 to 26.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-23

a method for introducing a foreign DNA of interest into a preselected site of a genome of a plant cell comprising the steps of
(a) inducing a double stranded DNA break at the preselected site in the genome of the cell ;
(b) introducing the foreign DNA of interest into the plant cell;
characterized in that the foreign DNA is delivered by direct DNA transfer

---

2. claims: 24-31

an isolated DNA fragment comprising a nucleotide sequence encoding the amino acid sequence of SEQ ID No 1 (I-SceI), wherein the nucleotide sequence has a GC content of about 50% to about 60%, and which meets the provisos recited in claim 24

International Application No
PCT/EP2004/013122

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12N15/82 C12N9/22

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, WPI Data, BIOSIS, Sequence Search, EMBL

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 03/054189 A (SUNGENE GMBH & CO. KGAA; BIESGEN, CHRISTIAN) 3 July 2003 (2003-07-03) the whole document	1-23
Y	page 18	27-31
Y	TZFIRA TZVI ET AL: "Site-specific integration of Agrobacterium tumefaciens T-DNA via double-stranded intermediates." PLANT PHYSIOLOGY (ROCKVILLE), vol. 133, no. 3, November 2003 (2003-11), pages 1011-1023, XP002319962 ISSN: 0032-0889 the whole document	1-23

-/--

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
14 April 2005	06 JUN 2005

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Bucka, A

6

Form PCT/ISA/210 (second sheet) (January 2004)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	PUCHTA H ET AL: "Two different but related mechanisms are used in plants for the repair of genomic double-strand breaks by homologous recombination" PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF USA, NATIONAL ACADEMY OF SCIENCE. WASHINGTON, US, vol. 93, May 1996 (1996-05), pages 5055-5060, XP002236852 ISSN: 0027-8424 page 5058, right-hand column page 5060, left-hand column	1-23
Y	CHILTON MARY-DELL M ET AL: "Targeted integration of T-DNA into the tobacco genome at double-stranded breaks: New insights on the mechanism of T-DNA integration." PLANT PHYSIOLOGY (ROCKVILLE), vol. 133, no. 3, November 2003 (2003-11), pages 956-965, XP002319963 ISSN: 0032-0889 the whole document	1-23
Y	PASZKOWSKI J ET AL: "GENE TARGETING IN PLANTS" EMBO JOURNAL, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 7, no. 13, 1988, pages 4021-4026, XP009001858 ISSN: 0261-4189 the whole document	1-23
A	PUCHTA H ET AL: "HOMOLOGOUS RECOMBINATION IN PLANT CELLS IS ENHANCED BY IN VIVO INDUCTION OF DOUBLE STRAND BREAKS INTO DNA BY A SITE-SPECIFIC ENDONUCLEASE" NUCLEIC ACIDS RESEARCH, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 21, no. 22, 11 November 1993 (1993-11-11), pages 5034-5040, XP000571587 ISSN: 0305-1048 the whole document	1-23
A	CHOULIKA A ET AL: "INDUCTION OF HOMOLOGOUS RECOMBINATION IN MAMMALIAN CHROMOSOMES BY USING THE I-SCEI SYSTEM OF SACCHAROMYCES CEREVISIAE" MOLECULAR AND CELLULAR BIOLOGY, WASHINGTON, DC, US, vol. 15, no. 4, 1 April 1995 (1995-04-01), pages 1968-1973, XP000572017 ISSN: 0270-7306 the whole document	1-23

-/--

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	KRYSIAK CEZARY ET AL: "Generation of DNA double-strand breaks and inhibition of somatic embryogenesis by tungsten microparticles in wheat" PLANT CELL TISSUE AND ORGAN CULTURE, vol. 58, no. 3, 1999, pages 163-170, XP002319964 ISSN: 0167-6857 the whole document	1-23
A	SALOMON S ET AL: "Capture of genomic and T-DNA sequences during double-strand break repair in somatic plant cells" EMBO JOURNAL, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 17, no. 20, 15 October 1998 (1998-10-15), pages 6086-6095, XP002259898 ISSN: 0261-4189 page 6093, right-hand column	1-23
A	SRIVASTAVA VIBHA ET AL: "Biolistic mediated site-specific integration in rice" MOLECULAR BREEDING, vol. 8, no. 4, 2001, pages 345-350, XP002320020 ISSN: 1380-3743 the whole document	1-23
Y	WO 96/14408 A (INSTITUT PASTEUR; UNIVERSITE PIERRE ET MARIE CURIE) 17 May 1996 (1996-05-17) page 13	27-31
Y	US 5 689 052 A (BROWN ET AL) 18 November 1997 (1997-11-18) column 4 - column 7	27-31
Y	FENNOY S L ET AL: "SYNONYMOUS CODON USAGE IN ZEA MAYS L. NUCLEAR GENES IS VARIED BY LEVELS OF C AND G-ENDING CODONS" NUCLEIC ACIDS RESEARCH, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 21, no. 23, 1993, pages 5294-5300, XP001122294 ISSN: 0305-1048 the whole document	27-31

-/--

International Application No
PCT/EP2004/013122

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	KAWABE AKIRA ET AL: "Patterns of codon usage bias in three dicot and four monocot plant species." GENES & GENETIC SYSTEMS, vol. 78, no. 5, October 2003 (2003-10), pages 343-352, XP002324073 ISSN: 1341-7568 the whole document	27-31
A	MURRAY E E ET AL: "CODON USAGE IN PLANT GENES" NUCLEIC ACIDS RESEARCH, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 17, no. 2, 25 January 1989 (1989-01-25), pages 477-498, XP000008653 ISSN: 0305-1048 the whole document	27-31

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 03054189 A	03-07-2003	AU 2002360986 A1	09-07-2003
		CA 2470329 A1	03-07-2003
		WO 03054189 A2	03-07-2003
		EP 1458875 A2	22-09-2004
WO 9614408 A	17-05-1996	US 5792632 A	11-08-1998
		CA 2203569 A1	17-05-1996
		WO 9614408 A2	17-05-1996
		EP 0791058 A1	27-08-1997
		JP 10508478 T	25-08-1998
		US 5948678 A	07-09-1999
		US 2003182670 A1	25-09-2003
		US 6822137 B1	23-11-2004
		US 5866361 A	02-02-1999
		US 6833252 B1	21-12-2004
		US 6395959 B1	28-05-2002
		US 2005032223 A1	10-02-2005
US 5689052 A	18-11-1997	US 6180774 B1	30-01-2001